Asian Journal of Biochemistry

ISSN 1815-9923

Academic
Journals Inc.

www.academicjournals.com

Asian Journal of Biochemistry 6 (4): 366-372, 2011
ISSN 1815-9923 / DOI: 10.3923/ajb.2011.366.372

Insulin Resistance and Interleukin-1β During Normal Pregnancy

[1]Abdolreza Sotoodeh Jahromi, [2]Parvin Zareian and [3]Abdolhossein Madani
[1]Department of Immunology, Jahrom University of Medical Sciences, Jahrom, Iran
[2]Department of Physiology, Jahrom University of Medical Sciences, Jahrom, Iran
[3]Department of Public Health, Hormozgan University of Medical Sciences, Bandarabbas, Iran

Corresponding Author: Parvin Zareian, Department of Physiology, Jahrom University of Medical Sciences, Jahrom, Iran
Tel: +98-791-3340406 Fax: +98-21-44670601

ABSTRACT

The purpose of this study was to evaluate the role of Interleukin-1β (IL-1β) in Insulin Resistance (IR) during normal pregnancy. This cross sectional study was carried out on 86 healthy pregnant women including 26, 23 and 37 individuals in the 1st, 2nd and 3rd trimesters, respectively and in 21 healthy non pregnant women. Serum IL-1β and resistin concentrations were measured by Enzyme Linked Immunosorbent Assay (ELISA) method. Insulin resistance value was calculated using the homeostasis model assessment, (Homeostasis Model Assessment) HOMA-IR. Serum resistin concentration was found to be significantly raised in the second and third trimester as compared with women with first trimester of pregnancy but it was not found any statistical difference in serum resistin concentration between the healthy controls and pregnant women. There was no difference in IL-1β level between pregnant 1.32±0.48 ng mL^{-1} and non pregnant women 1.44±0.49 ng mL^{-1}. IL-1β level were significantly decreased with increase in gestational age. Pregnant women exhibited higher score of HOMA-IR 2.1±0.9 compared non pregnant women 1.7±0.4 but there were not difference in this score between pregnant subjects in different gestational age. There was not any correlation between maternal gestational age and serum level of IL-1β. The findings of this study suggest that IL-1β do not appear to contribute greatly to pregnancy induced insulin resistance in healthy pregnancy.

Key words: Insulin, resistin, interleukin-1β, gestation, HOMA-IR

INTRODUCTION

Metabolic Syndrome (MS) is a collection of risk factors including insulin resistance, central obesity, hypertension and dyslipidemia and itself is a risk factor for coronary artery disease (Mahajan *et al*., 2010). Pregnancy is related to glucose metabolism disorders represents a state of Insulin Resistance (IR) (Hadden and McLaughlin, 2009; Johnson, 2008).

Insulin resistance may facilitate supply of appropriate nutrients particularly of glucose to fetus for fetal growth and metabolism. The mechanism responsible for insulin resistance has not been clearly stated. Insulin resistance can be affected by Nitric Oxide (NO) (McGrowder and Brown, 2007). Recent researches have been shown adipokinins include leptin (Mohiti *et al*., 2009; Assal *et al*., 2007; El-Ghaffar and El-Said, 2006), resistin (Caja *et al*., 2005), TNf-α (Kirwan *et al*., 2002) and cytokines include IL-6 and IL-1β (Nov *et al*., 2010) play an important role in insulin resistance. Insulin sensitivity changes from an enhanced state during early pregnancy to an insulin resistant state in late pregnancy. Insulin sensitivity changes from an enhanced state during early pregnancy to an insulin resistant state in late pregnancy (Kirwan *et al*., 2002).

Therefore, it is inspected, subsequent to increase in IR during pregnancy, its related factors changes, too. In a recently published study, it has been reported that in normal pregnant women both maternal IL-6 and TNF-α cytokines are increased but we could not find any correlation between IR with IL-6 and TNF-α (Jahromi *et al*., 2011).

IL-1β is one of the cytokines produce by immune and non immune cells (Maedler *et al*., 2009; Soliman *et al*., 2009). This cytokine has a role in response to inflammation, infection or injury (Yang *et al*., 2004). In addition, there are reports regarding its role in the onset of normal labor (Steinborn *et al*., 1999). A few studies suggest that IL-1β induce insulin resistance in adipocytes and hepatocytes (Nov *et al*., 2010; Jager *et al*., 2007). Resistin is an insulin-sensitizing adipokine that can be implicated in endogenous glucose regulation (Mohiti *et al*., 2009).

However, its correlation with IL-1β in insulin resistance during normal pregnancy has not been studied. Therefore, this study has been aimed to evaluate whether serum IL-1β concentration changes during normal pregnancy and their correlation with known markers of IR such as Resistin and BMI.

MATERIALS AND METHODS

Setting and sample: This study was conducted at the department of Obstetrics and Gynecology of Honary Clinic, Jahrom, Iran, 2010. Subjects were 86 pregnant women with different gestational ages (first trimester: 26, second trimester: 23, third trimester: 37 and 21 non pregnant women similar in age and Body Mass Index (BMI). All subjects met the following criteria: no history of pre- gestational diabetes; no history of liver, respiratory, thyroid or other illness and any current infectious condition. They were not on any drug therapy.

Body mass index calculation: Body Mass Index (BMI, Kg m^{-2}) was calculated according to the maternal height and pre-pregnancy weight (Hammer *et al*., 1991).

Determination of blood sugar: Blood sugar was measured by glucose oxidase/peroxidase (GOD-POD) method (Maughan, 1982).

Determination of serum level of IL-1β and Insulin: Serum samples were analysed for concentrations of IL-1β, insulin by Enzyme Linked Immunosorbent Assay (ELISA) (Anderson *et al*., 1993).

Serum insulin was determined by ELISA (Diaplus; based on the direct sandwich technique in which two monoclonal antibodies are directed against separate antigenic determinants on the insulin molecule. During incubation insulin in the sample reacts with enzyme (HRP) conjugated anti-insulin antibody and anti-insulin antibody bound to micro-titration well. A sample washing step removed unbound enzyme labeled antibody. In the insulin ELISA, the bound HRP complex is detected by reaction with TMB substrate. The reaction is stopped by adding acid to give a colorimetric endpoint that is read using ELISA reader).

Serum IL-1β was measured by ELISA (Bendermed, Austria: Cat. No. BMS224INSTCE). An anti-human IL-1β coating antibody is adsorbed onto micro wells. Human IL-1β present in the sample or standard binds to antibodies adsorbed to the micro wells; a biotin-conjugated monoclonal anti-human IL-1β antibody binds to human IL-1β captured by the first antibody. Streptavidin-HRP binds to the biotin conjugated anti-human IL-1β. Following incubation unbound

biotin conjugated anti human IL-1β and Streptavidin-HRP is removed during a wash step and substrate solution reactive with HRP is added to the wells. A colored product is formed in proportion to the amount of soluble human IL-1β present in the sample. The reaction is terminated by addition of acid and absorbance is measured at 450 nm. A standard curve is prepared from seven human IL-1β standard dilutions and human IL-1β sample concentration determined.

Determination of insulin resistance: Insulin resistance value were calculated using the homeostasis model assessment, HOMA- IR ,as (fasting insulin IU/L) × (fasting glucose mmol L^{-1}) /22.5 as previously reported by Matthews *et al.* (1985).

Statistical analysis: All results are displayed as Mean±SD (standard deviation of mean) and Min-Max. Resistin, Insulin Resistance (IR), BMI and body weight data were analyzed with One Way Analysis of Variance (ANOVA). Serum IL-1β and insulin concentration data were analyzed with non-parametric kruskal-wallis test (Vargha and Delaney, 1998) followed by Mann Whitney U-test (Rosner and Grove, 1999). Correlations were calculated using liner correlation (pearson). Statistical analysis was performed using SPSS 11 for windows. $p<0.05$ was considered statistically significant for all analysis.

RESULTS

A total of 86 pregnant women and 21 non pregnant subjects participated in the study. Clinical and laboratory characteristics of pregnant women and controls are summarized in Table 1. There were not significant differences in IL-1β level, resistin level and insulin level in pregnant as compared with controls.

BMI were found to be significantly increased in the 3rd trimester (17.26-34.1) kg m^{-2} as compared with controls (18.4-31.1) kg m^{-2} and women with 1st trimester of pregnancy (20.2-32.9) kg m^{-2} (Table 2).

Pregnant women in the second (100-130) mmHg and third trimester (100-130) mmHg of pregnancy had significantly higher systolic pressure than non pregnant women (100-120) mmHg.

Serum resistin concentration were found to be significantly raised in the second (5.9-14.3) ng mL^{-1} and third (1.23-15.8) ng mL^{-1} trimester as compared with women with first trimester (4.6-8.2) ng mL^{-1} of pregnancy but it was not found any statistical difference in serum resistin concentration between the healthy controls and pregnant women with gestational age less than 24 weeks (Table 2).

There was no difference in IL-1β level between pregnant (0.47-2.65) ng mL^{-1} and non pregnant (0.7-2.35) ng mL^{-1} women (Table 1). IL-1β level were significantly decreased with increase in gestational age (Table 2). Pregnant women exhibited higher score of HOMA IR (0.57-3.97) compared non pregnant women (0.98-2.4) but there were not difference in this score between pregnant subjects in different gestational age (Table 1, 2). There were significant correlation between gestational age and BMI ($r = 0.28$, $p = 0.01$), diastolic pressure ($r = 0.28$, $p = 0.01$) resistin ($r = 0.36$, $p = 0.002$) and IL-1β level ($r = -0.45$, $p<0.000$). There was not significant correlation between gestational age and IR.

Resistin level in pregnancy did not correlate with IR, fasting insulin, BMI and body weight. IL-1β level also did not correlate with IR, fasting insulin, BMI and body weight.

Table 1: Clinical and laboratory characteristic of patients and control

Characteristic	Control	Pregnant women
No. of case	86	21
Age (year)	26.4±4.19(18-38)	27.2±5.6 (20-41)
Gestational age (Week)	23.9±9.8 (8-40)	
HT (m)	1.6±0.06 (1.45-1.8)	1.58±0.07 (1.5-1.7)
WT (kg)	64.99±11.5[b] (45-88)	58.6±6.4 (49-70)
BMI (kg m^{-2})	25.4±3.7[a] (17.26-34.13)	23.4±3 (18.4-31.1)
SBP (mmHg)	117±7.8[b] (100-130)	110.7±5.9 (100-120)
DBP (mm Hg)	72.8±6.9 (60-80)	70.9±11.4 (60-100)
BGL (mg 100^{-1})	81.5 ±8.7 (62-194)	80.2±8.7 (65-97)
Insulin (μLU mL^{-1})	10.9±6 (2.8-54.1)	8.7±1.9 (4.2-11.7)
Resistin (ng mL^{-1})	8.3±2.6 (1.23-15.8)	7±3.3 (2.9-14.3)
IL-1β (ng mL^{-1})	1.32±0.48 (0.47-2.65)	1.44± 0.49 (0.7-2.35)
IR	2.1±0.9 (0.57-3.97)	1.7±0.4 (0.98-2.4)

Value are Mean±SD (min-max). BMI: Body max index, HT: Height of women, WT: Weight of body, SBP: Systolic blood pressure, DBP: Diastolic blood pressure, BGL: Blood glucose level, IR: Insulin resistance, IL-1β: Interleukin -1 beta, [a]$p<0.05$ (control), [b]$p<0.01$ (control)

Table 2: Clinical and laboratory characteristics of pregnant women with different gestational age

Characteristic	1st trimester	2nd trimester	3rd trimester
No. of cases	26	23	37
Age (year)	25.5±4.1 (18-36)	24.9±2.3 (20-29)	27.6±4.7 (21-38)
GA (week)	11.2±1.6 (8-14)	22.2±2.7 (15-25)	32.9±4.3 (26-40)
WT (kg)	59.2±10.2 (47-77)	65.7±12.6 (45-88)	68.2±10.6[b] (45-88)
HT (m)	1.58±0.06 (1.5-1.7)	1.58±0.06 (1.5-1.7)	1.6±0.07 (1.45-1.8)
SBP (mm Hg)	114.2±8.7 (100-130)	117.5±7.7 (100-130)	118.3±7[a] (100-130)
DBP (mm Hg)	70.5±6.6 (60-80)	71.7±7.1 (60-80)	74.7±6.5 (60-80)
BMI (kg m^{-2})	23.6±3.6 (20.2-32.9)	25.7±3.7 (20.4-33.7)	26.2±3.6[b] (17.26-34.1)
BGL (mg 100^{-1})	78.6±6.7 (67-88)	79.4±6.7 (62-105)	84.1±21.2 (63-194)
Insulin (μL mL^{-1})	10.5±3.4 (5.8-19.4)	10.6±3.8 (2.8-54.1)	10.05±3.9 (3.1-18)
Resistin (ng mL^{-1})	6.7±0.97 (4.6-8.2)	8.6±2[a] (5.9-14.3)	9.5±3.3[c] (1.23-15.8)
IL-1β	1.59±0.38 (1.05-2.4)	1.26±0.41[a] (0.75-2.1)	1.07±0.42[c] (0.5-2.7)
IR	2±0.6 (0.98-3.27)	2.02±0.66 (1.05-3.56)	2.1±1.6 (0.57-3.97)

Values are mean±SD (min-max). BMI: Body mass index, GA: Gestational age, WT: Wight of body, HT: Height, SBP: Systolic blood pressure, DBP: Diastolic blood pressure, BGL: Blood glucose level, IR: Insulin resistance, IL-1β: Interleukin-1 beta, [a]$p<0.05$, [b]$p<0.01$, [c]$p<0.001$ (Significantly different from pregnant women in 1st trimester)

DISCUSSION

Glucose metabolism disorder is a common complication during pregnancy and its pathology is associated with IR and deficiency of insulin secretion. In this study, insulin resistance significantly was higher in total group of healthy pregnant than in non-pregnant women. In spite of recent previous study (Jahromi *et al*., 2011) it was not found correlation between gestational age and insulin resistance in present study. It has been shown by Kirwan that insulin resistance was significantly increased in late pregnancy compared with either control or early pregnancy (Kirwan *et al*., 2002). The results of these studies are in contradiction to the results of present study. This difference may be related to differences in dietary composition, life style between western and eastern societies (Clapp, 2006; Sivabalan and Menon, 2008), variability between insulin assays in different experimental researches, differences in the population studied and sampling time during pregnancy (Manley *et al*., 2008).

Cytokine proteins classifies base on their T-cell linage as helper T lymphocyte 1 (Th1) or helper T lymphocyte 2 (Th2). There are reports demonstrated that in normal pregnant women circulating Th2 cell-derived cytokines increased and Th1 cell derived cytokines decreased (Marzi *et al*., 1996). An increasing in both maternal IL-6 (Th2) and TNF –α (Th1) cytokines was reported that in normal pregnant women (Jahromi *et al*., 2011).

In this study maternal IL-1β concentration in pregnant women is lower than non pregnant women and showed a significant negative correlation between IL-1β and gestational age. Maternal serum IL-1β decreased with further increase in pregnancy period. The present study is the first to report maternal IL-1β concentration during normal pregnancy. AS IL-1β is participate in immune system parallel to Th1 cells (Sobhani *et al*., 2008) and it is categorized as a Th1 cytokine and as Th1 cytokines are decreased in pregnancy, thus the results of present study is support by previous research (Szarka *et al*., 2010; Jahromi *et al*., 2011).

In an *in vitro* study, measurement of IL-1β concentration in the 3rd trimester has been shown that IL-1β production was unchanged (Russella *et al*., 1997). The result of this study is in contradiction to the result of present study. This difference may be related to differences in genetics constitution (Lee *et al*., 2008), variability between insulin assays in different experimental researches, differences in the population studied and sampling time during pregnancy (Manley *et al*., 2008). It seems, there is a gradual decline in the expression of IL-1β as trophoblastic cells progressively differentiate into syncytial cells (Das *et al*., 2002).

According to the findings of present investigation, "it was not found correlation between maternal IL-1β and insulin resistance". It can be concluded that IL-1β do not appear to contribute greatly to pregnancy induced insulin resistance in healthy pregnant women.

REFERENCES

Anderson, L., B. Dinesen, P.N. Jorgesen, F. Poulsen and M.F. Roder, 1993. Enzyme immunoassay for intact human insulin in serum or plasma. Clin. Chim. Acta., 39: 578-585.

Assal, H.S., M. Fath-Allah and A. Elsherbiny, 2007. Serum leptin and adiponectin in obese diabetic and non-diabetic. J. Medical Sci., 7: 865-869.

Caja, S., I. Martinez, M. Abelenda and M. Puerta, 2005. Resistin expression and plasma concentration peak at different times during pregnancy in rats. J. Endocrinol., 185: 551-559.

Clapp, J.F., 2006. Effects of diet and exercise on insulin resistance during pregnancy. Metab. Syndr. Relat. Disord., 4: 84-90.

Das, C., V.S. Kumar, S. Gupta and S. Kumar, 2002. Network of cytokines, integrins and hormones in human trophoblast cells. J. Reprod. Immunol., 53: 257-268.

El-Ghaffar, N.A. and N.H. El-Said, 2006. Hypoadiponectinemia in Egyptian patients with type ii diabetes mellitus with vascular complications. J. Med. Sci., 6: 626-630.

Hadden, D.R. and C. McLaughlin, 2009. Normal and abnormal maternal metabolism during pregnancy. Semin Fetal Neonatal Med., 14: 66-71.

Hammer, L.D., H.C. Kraemer, D.M. Wilson, P.L. Ritter and S.M. Dornbusch, 1991. Standardized percentile curves of body-mass index for children and adolescents. Am. J. Dis. Child., 145: 259-263.

Jager, J., T. Gremeaux, M. Cormont, Y.L. Marchand-Brustel and J.F. Tanti, 2007. Interleukin-1β-induced insulin resistance in adipocytes through down-regulation of insulin receptor substrate-1 expression. Endocrinology, 148: 241-251.

Jahromi, A.S., P. Zareian and A. Madani, 2011. Association of insulin resistance with serum interleukin-6 and TNF-α levels during normal pregnancy. Biomarker Insights, 6: 1-6.

Johnson, C.A., 2008. Glucose homeostasis during canine pregnancy: Insulin resistance, ketosis and hypoglycemia. Theriogenology, 70: 1418-1423.

Kirwan, J.P., S.H.D. Mouzon, J. Lepercq, J.C. Challier and L. Huston-Presley *et al.*, 2002. TNF-α is a predictor of insulin resistance in human pregnancy. Diabetes, 51: 2207-2213.

Lee, J.H., Y.D. Kwon, S.H. Hong, H.J. Jeong, H.M. Kim and J.Y. Um, 2008. Interleukin-1 beta gene polymorphism and traditional constitution in obese women. Int. J. Neurosci., 118: 793-805.

Maedler, K., G. Dharmadhikari, D.M. Schumann and J. Storling, 2009. Interleukin-1 beta targeted therapy for type 2 diabetes. Expert Opin. Biol. Ther., 9: 1177-1188.

Mahajan, R., K. Gupta and V. Kapoor, 2010. A systematic account of pathogenesis, diagnosis and pharmacotherapy of metabolic syndrome: Things we need to know. Int. J. Pharmacol., 6: 338-345.

Manley, S.E., S.D. Luzio, I.M. Stratton, T.M. Wallace and P.M. Clark, 2008. Preanalytical, analytical and computational factors affect homeostasis model assessment. Diabetes Care, 31: 1877-1883.

Marzi, M., A. Vigano, D. Trabattoni, M.L. Villa, A. Salvaggio, E. Clerici and M. Clerici, 1996. Characterization of type 1 and type 2 cytokine production profile in physiologic and pathologic human pregnancy. Clin. Exp. Immunol., 106: 127-133.

Matthews, D.R., J.P. Hosker, A.S. Rudenski, B.A. Naylor, D.F. Treacher and R.C. Turner, 1985. Homeostasis model assessment: Insulin resistance and β-cell function from fasting plasma glucose and insulin concentration in man. Diabetologia, 28: 412-419.

Maughan, R.J., 1982. A simple, rapid method for the determination of glucose, lactate, pyruvate, alanine, 3-hydroxybutyrate and acetoacetate on a single 20-mul blood sample. Clin. Chim. Acta, 122: 231-240.

McGrowder, D. and P.D. Brown, 2007. Effects of nitric oxide on glucose transport: *In vivo* and *in vitro* studies. Asian J. Biochem., 2: 1-18.

Mohiti, J., F. Talebi and M. Afkhami-Ardekani, 2009. Circulation free leptin in diabetic patients and its correlation to insulin level. Pak. J. Biol. Sci., 12: 397-400.

Nov, O., A. Kohl, E.C. Lewis, N. Bashan and I. Dvir *et al.*, 2010. Interleukin-1β may mediate insulin resistance in liver-derived cells in response to adipocyte inflammation. Endocrinology, 151: 4247-4256.

Rosner, B. and D. Grove, 1999. Use of the Mann-Whitney U-test for clustered data. Stat. Med., 18: 1387-1400.

Russella, A.S., C. Johnston, C. Chew and W.P. Maksymowych, 1997. Evidence for reduced Th1 function in normal pregnancy: A hypothesis for the remission of rheumatoid arthritis. J. Rheumatol., 24: 1045-1050.

Sivabalan, S. and V.P. Menon, 2008. Effect of feeding high fat with or without sucrose on the development of diabetes in wistar rats. Asian J. Biochem., 3: 271-279.

Sobhani, M.E., N. Haque, A.T.M.K. Islam, U. Salma, A. Ahmed, I.J. Mukti and A.K.M.F. Haque, 2008. Brief naturalistic stressors cause shift of TH1 to TH2 cytokine response and increase disease susceptibility. Biotechnology, 7: 623-629.

Soliman, M.M., Y.A. El-Fattah El-Senosi, O.M.A. El-Hamid, A.El-Desouki Abd El-Mageed, R.S. Ismaeil and H.A. El-Maqsoud Ali, 2009. *Nigella sativa* modulates cytokines expression in mature bovine adipocytes. Asian J. Biochem., 4: 60-67.

Steinborn, A., A. Niederhut, C. Solbach, R. Hildenbrand, C. Sohn and M. Kaufmann, 1999. Cytokine release from placental endothelial cells, a process associated with preterm labour in the absence of intrauterine infection. Cytokine, 11: 66-73.

Szarka, A., J. Rigo, Jr., L. Lazar, G. Beko and A. Molvarec, 2010. Circulating cytokines, chemokines and adhesion molecules in normal pregnancy and preeclampsia determined by multiplex suspension array. BMC Immunol., 2: 11-59.

Vargha, A. and H.D. Delaney, 1998. The Kruskal-Wallis test and stochastic homogeneity. J. Educ. Behav. Stat., 23: 170-192.

Yang, X.P., U. Albrecht, V. Zakowski, R.M. Sobota and D. Haussinger *et al.*, 2004. Dual function of interleukin-1beta for the regulation of interleukin-6-induced suppressor of cytokine signaling 3 expression. J. Biol. Chem., 279: 45279-45289.